Table II

Physical characteristics of 3'-*O*-(2-methoxyethyl) containing 2'-5' linked oligonucleotides.

	Expected Mass	Observed Mass	HPLC[2] T_R Purified (min.)	#Ods(260nm)
17176	6440.743	6440.300	23.47	3006
17177	6514.814	6513.910	23.67	3330
17178	6482.814	6480.900	23.06	390
17179	6513.798	6513.560	23.20	3240
17180	6588.879	6588.200	23.96	3222
17181	6540.879	6539.930	23.01	
21415	6662.976	6662.700	24.18	4008
21416	6598.969	6597.800	23.01	3060
21945	1099.924	1099.300	19.92	121
21663	1487.324	1486.800	20.16	71
20389	1483.000	1482.000		62
20390	4588.000	4591.000		151

[2]Conditions: Waters 600E with detector 991; Waters C4 column (3.9X300mm); Solvent A: 50 mM TEA-Ac, pH 7.0; B: 100% acetonitrile; 1.5 mL/min. flow rate; Gradient: 5% B for first five minutes with linear increase in B to 60% during the next 55 minutes.

EXAMPLE 51

T_m Studies on modified oligonucleotides

[0188] Oligonucleotides synthesized in Examples 49 and 50 were evaluated for their relative ability to bind to their complementary nucleic acids by measurement of their melting temperature (T_m). The melting temperature (T_m), a characteristic physical property of double helices, denotes the temperature (in degrees centigrade) at which 50% helical (hybridized) versus coil (unhybridized) forms are present. T_m is measured by using the UV spectrum to determine the formation and breakdown (melting) of the hybridization complex. Base stacking, which occurs during hybridization, is accompanied by a reduction in UV absorption (hypochromicity). Consequently, a reduction in UV absorption indicates a higher T_m. The higher the T_m, the greater the strength of the bonds between the strands.

[0189] Selected test oligonucleotides and their complementary nucleic acids were incubated at a standard concentration of 4 μM for each oligonucleotide in buffer (100 mM NaCl, 10 mM sodium phosphate, pH 7.0, 0.1 mM EDTA). Samples were heated to 90 °C and the initial absorbance taken using a Guilford Response II Spectrophotometer (Corning). Samples were then slowly cooled to 15 °C and then the change in absorbance at 260 nm was monitored with

heating during the heat denaturation procedure. The temperature was increased by 1 degree °C/absorbance reading and the denaturation profile analyzed by taking the 1st derivative of the melting curve. Data was also analyzed using a two-state linear regression analysis to determine the Tm=s. The results of these tests for the some of the oligonucleotides from Examples 49 and 50 are shown in Table III below.

Table III

Tm Analysis of Oligonucleotides

SEQ ID: NO. #	(ISIS)#	Sequence (5'-3')	Backbone	T_m 8	# Mods	#2'-5' Linkages
13	(11061)	ATG-CAT-TCT-GCC-CCC-AAG-GA	P=S	61.4	0	0
4	(17176)	ATG-CAT-TCT-GCC-CCC-AAG-GA*	P=S	61.4	1	0
5	(17177)	ATG-CAT-TCT-GCC-CCC-AAG-**G*A***	P=S	61.3	2	1
6	(17178)	ATG-CAT-TCT-GCC-CCC-AAG$_O$-**G$^*_O$A***	P=S/P=O	61.8	2	1
7	(17179)	A*TG-CAT-TCT-GCC-CCC-AAG-GA*	P=S	61.1	2	1
8	(17180)	A*TG-CAT-TCT-GCC-CCC-AAG-**G*A***	P=S	61.0	3	2
9	(17181)	A$^*_O$TG-CAT-TCT-GCC-AAA-AAG$_O$-**G$^*_O$A***	P=S/P=O	61.8	3	2
10	(21415)	**A*T***G-CAT-TCT-GCC-AAA-AAG-**G*A***	P=S	61.4	4	3
11	(21416)	**A$^*_O$T$^*_O$**G-CAT-TCT-GCC-AAA-AAG$_O$-**G$^*_O$A***	P=S/P=O	61.7	4	3